= 7.1 Hz, 28-methyl), 0.68 (3H, s, 18-methyl); MS *m/e*: 416 (M^+), 398 ($M^+ - H_2O$), 383, 365, 314, 273, 271, 87, 85, 83, 81, 59, 57, 55.

Physalindicanol B monoacetate (**2b**)

A solution of physalindicanol B (10 mg) in pyridine (1 ml) and Ac_2O (0.5 ml) was left overnight under anhydrous condition at room temperature. After usual work up followed by crystallisation from MeOH, the monoacetate (**2b**) was obtained as needles (7 mg), m.p. 155–157° C; IR ($CHCl_3$) ν_{max} cm^{-1}: 3592, 2980, 1720, 1517, 1468, 1420, 1145, 927; 270 MHz ^{1}H-NMR ($CDCl_3$): 5.37 (1H, d, J = 5.2 Hz, H-6), 5.09 and 4.76 (1H, s, each, H_2-28), 4.56 (1H, septet, H-3), 2.03 (3H, s, OAc), 1.35 (6H, s, 26- and 27-methyls), 1.02 (3H, s, 19-methyl), 0.97 (3H, d, J = 6.5 Hz, 21-methyl), 0.69 (3H, s, 18-methyl); MS *m/e*: 456 (M^+), 396 ($M^+ - AcOH$), 381, 378, 363, 314, 312, 296, 271, 85, 81, 59, 57, 55.

Acknowledgements

Sincere thanks are due to Prof. Dr. Lavie, The Weizmann Institute of Science, Rehovot, Israel and to Dr. K. N. Garg, Rohtak Medical College, India for the ^{1}H-NMR and IR spectra of ergosta-5,25-dien-3β,24-diol, isolated from *W. coagulans*. The authors are also thankful to Prof. Paul L. Schiff, Jr., School of Pharmacy, University of Pittsburgh, U.S.A., Dr. M. N. Khanna, CDRI, Lucknow and Dr. S. C. Pakrashi, Institute of Chemical Biology, Calcutta for various spectral data. One of us (S.C.S.) is grateful to C.S.I.R., New Delhi, for financial assistance.

References

(1) The Wealth of India, (1969) A Dictionary of Indian Raw Materials & Industrial Products, Vol. VIII, p. 38, C.S.I.R. Publication, New Delhi.
(2) Chopra, R. N., Nayar, S. L., Chopra, I. C. (1956) Glossary of Indian Medicinal Plants, p. 192, C.S.I.R. Publication, New Delhi.
(3) Kirtikar, K. R., Basu, B. D. (1975) Indian Medicinal Plants, Vol. III, p. 1768, Singh & Singh Publication, Dehradun.
(4) Ali, A., Sahai, M., Ray, A. B., Slatkin D. J. (1984) J. Nat. Prod. 47, 648 and references cited therein.
(5) Bagchi, A., Neogi, P., Sahai, M., Ray, A. B., Oshima, Y., Hikino, H. (1984) Phytochemistry 23, 853.
(6) Sahai, M., Kirson, I. (1984) J. Nat. Prod. 47, 527.
(7) Velde, V. V., Lavie, D., Budhiraja, R. D., Sudhir, S., Garg, K. N. (1983) Phytochemistry 22, 2253.
(8) Glotter, E., Kirson, I., Lavie, D., Abraham, A. (1978) in Bioorganic Chemistry, (van Tamelen, E. E., ed.) Vol. II, Ch. 3, Academic Press, New York.
(9) Kirson, I., Glotter, E. (1981) J. Nat. Prod. 44, 633.
(10) Bhacca, N. S., Williams, D. H. (1964) Application of NMR Spectroscopy in Organic Chemistry, pp. 80, 181. Holden-Day, San-Fransisco.
(11) Bridgeman, J. E., Cherry, P. C., Clegg, A. S., Erans, J. M., Jones Ewart, R. H., Kasal, A., Kumar, V., Meakins, G. D., Morisawa, Y., Richards, E. E., Woodgate, P. D. (1970) J. Chem. Soc. (C), 250.
(12) Wyllie, S. G., Djerassi, C. (1968) J. Org. Chem. 33, 305.
(13) Iribarren, A. M., Pomillo, A. B. (1983) J. Nat. Prod. 46, 752.
(14) Lockley, W. J. S., Rees, H. H., Goodwin, T. W. (1976) Phytochemistry 15, 937.
(15) The chemical shift of H-3 of the sterol isolated from *Withania coagulans* (Ref. No. 7) was erroneously reported by Velde et al. as δ = 3.29 ppm instead of δ = 3.51 ppm (personal communication from Prof. D. Lavie, The Weizmann Institute of Science, Rehovot, Israel).

Indole Alkaloids from *Trachelospermum jasminoides*

Atta-ur-Rahman[1,3], Talat Fatima[1], Mehrun-Nisa[1], S. Ijaz[1], George Crank[2] and Shaheen Wasti[2]

Received: April 8, 1986

Abstract: The leaves and stems of *T. jasminoides* have been found to contain indole alkaloids. Five indole alkaloids, coronaridine, voacangine, apparicine, conoflorine, and 19-epi-voacangarine have been isolated. The ^{13}C-NMR spectra of apparicine and 19-epi-voacangarine are also reported.

Introduction

T. jasminoides (Apocynaceae) is a glabrous evergreen tree, commonly growing in the gardens of India and Pakistan. Various parts of the plant are used in the indigenous system of medicine for the treatment of rheumatism, gonorrhea, carcinomatous growth, sciatica, viper bite, and wounds. The seeds are hemostatic, and cardiotonic (1). No alkaloidal constituents have previously been reported from this plant.

Results and Discussion

The crude alkaloids were isolated from the concentrated alcoholic extracts of the leaves and stems of *T. jasminoides* by extraction at different pH values. The fraction obtained at pH1 was subjected to flash column chromatography. The fraction eluted with petroleum ether : $CHCl_3$ (1 : 1) was found to contain 2 alkaloids. The minor one was identified as coronaridine (**1**) by comparison of its spectral data (UV, IR, MS, ^{1}H-NMR) with those reported in the literature (2), while the major one was identified as voacangine (**2**) by comparison of its spectral data (3) as well as by direct comparison with authentic sample. Voacangine was also isolated in low yields from the subsequent extraction at pH5. Further elution of the same column with, $CHCl_3$: MeOH (95 : 5), gave another major alkaloid which was identified as apparicine (**3**) by direct comparison with an authentic samples as well as by comparison of its spectral data with those reported in the literature (4). The ^{13}C-NMR data of apparicine (75 MHz, $CDCl_3$) are shown in Table I.

The fraction obtained at pH5 (20 g, F_2) was also chromatographed on a silica column. The fraction obtained on elution

---

[1] H. E. J. Research Institute of Chemistry, University of Karachi, Karachi-32, Pakistan.
[2] School of Chemistry, University of New South Wales, Sydney, Australia.
[3] Address for correspondence.

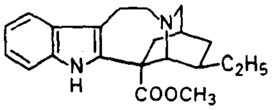
Table I. ^{13}C-NMR ($CDCl_3$) data for apparicine (**3**) and 19-epi-voacangarine (**5**)

Carbon No	Apparicine (3) Chemical shift (ppm)	19-epi-Voacangarine (5) Chemical shift (ppm)
2	144.72	134.60
3	45.16	52.32
5	–	50.99
6	53.78	21.63
7	112.64	109.84
8	129.00	130.71
9	118.61	100.93
10	119.55	154.35
11	121.35	111.28
12	123.15	112.43
13	136.62	136.30
14	29.08	26.98
15	41.14	28.58
16	135.80	57.53
17	109.88	36.77
18	12.64	22.25
19	110.39	70.64
20	131.60	40.37
21	54.16	54.35
$O\underline{C}H_3$	–	56.10
$C\overline{O}O\underline{C}H_3$	–	52.84
$\underline{C}OO\overline{C}H_3$	–	167.80

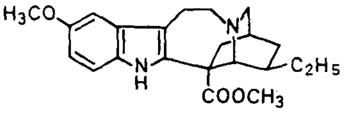
Multiplicity confirmed by gated spin echo measurements.

with petroleum ether: $CHCl_3$ (2:8) consisted of a mixture of two alkaloids. The slower moving alkaloid which was the major one, was identified by spectroscopic comparison (UV, IR, MS, ^1H-NMR) as conoflorine (**4**) (5).

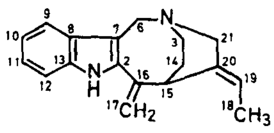
Further elution of the same column with $CHCl_3$:EtOAc (9:1) afforded another major alkaloid, identified by means of its spectral data as 19-epi-voacangarine (**5**) (6). The ^{13}C-NMR data of 19-epi-voacangarine (75 MHz, $CDCl_3$) are shown in Table I.



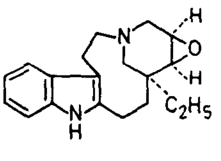
1 coronaridine



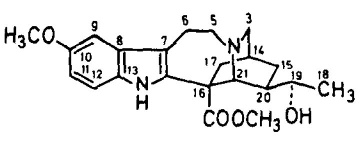
2 voacangine



3 apparicine



4 conoflorine



5 19-epi-voacangarine

Materials and Methods

Leaves and stems of *T. jasminoides* were collected from gardens Karachi University in January 1985. The plant was identified by Dr. I. Ali, Professor of Botany, University of Karachi, and a voucher spe men is deposited at the Department of Botany, University of Karac Pakistan.

Apparatus: Spectra was recorded on Jasco-IRA-1, IR spe trophotometer, Shimadzu UV-240, UV spectrophotometer, Finniga MAT 312 mass spectrometer linked to DEC-PDP 11/34 data system and Bruker AM-300 MHz NMR spectrometer.

Isolation of Alkaloids

Leaves and stems (50 kg) were dried in the shade and extracted wit EtOH. The alcoholic extracts were concentrated and partitioned be tween 10 % HCl and $CHCl_3$. The $CHCl_3$ layer was dried (Na_2SO_4) an evaporated to a gum (25 g; F_1). The aqueous acidic layer was basifie with aqueous NH_3 and extracted into $CHCl_3$ at various pH values. Th fraction obtained at pH5 (20 g, F_2) contained the major alkaloids.

The fraction (F_1, 25 g) was subjected to flash chromatography. The column was eluted with increasing polarities of mixtures of petroleum ether, $CHCl_3$, EtOAc, and MeOH. The fraction obtained on elution with petroleum ether: chloroform (1:1) was collected, and the eluates were concentrated. The fraction contained 2 alkaloids, which were separated by preparative TLC on silica gel plates (petroleum ether, acetone, ammonia, 9:1:0.05). The faster moving minor alkaloid (5 mg) was identified as coronaridine (**1**) while the slower one was identified as voacangine (**2**) (40 mg).

Coronaridine (**1**)

UV (MeOH): λ_{max}, 225, 285 and 294 nm; IR ($CHCl_3$): v, 1720 cm^{-1} (ester carbonyl); MS: *m/z* (%) = 338 (20), 323 (8), 309 (8), 279 (7), 253 (6), 214 (56), 148 (26), 136 (80), 124 (50), 122 (56); ^1H-NMR ($CDCl_3$): δ, 0.88 (t, 3H, $J_{18,19} = 6.9$ Hz, C-18H), 1.5 (m, 2H, C-19H), 3.72 (s, 3H, $COOC\underline{H}_3$), 7.09–7.48 (m, 4H, Ar-H), 7.75 (br. s, 1H, $N\underline{H}$).

Voacangine (**2**)

UV (MeOH): λ_{max}, 225, 285, 300 nm; IR ($CHCl_3$): v, 1630, 1720 cm^{-1}; MS: *m/z* (%) = 368 (20), 353 (10), 339 (8), 309 (12), 283 (14), 244 (26), 225 (12), 184 (30), 160 (25), 148 (16), 136 (100), 124 (34), 122 (36); ^1H-NMR ($CDCl_3$) δ, 0.89 (t, 3H, $J_{18,19} = 7.5$ Hz, C-18H), 1.27 (m, 2H, C-19H), 3.72 (s, 3H, $COOC\underline{H}_3$), 3.85 (s, 3H, Ar-$OC\underline{H}_3$), 6.81 (dd, 1H, $J_{11,12} = 8.7$ Hz, $J_{11,9} = 2.4$ Hz, C-11H), 6.93 (d, 1H, $J_{9,11} = 2.4$ Hz, C-9H), 7.19 (d, 1H, $J_{12,11} = 8.7$ Hz, C-12H), 7.75 (br. s, 1H, $N\underline{H}$).

Further elution of the same column with $CHCl_3$: MeOH (95:5) gave another major alkaloid (50 mg) which was purified by preparative TLC ($CHCl_3$: MeOH, 8:2). This alkaloid was identified as apparicine (**3**) (20 mg).

Apparicine (**3**)

UV (MeOH): λ_{max} 292, 303, 312 nm; IR ($CHCl_3$): v, 3350 cm^{-1}; MS: *m/z* (%) = 264 (100), 249 (28), 235 (34), 222 (51), 208 (77), 154 (42); ^1H-NMR ($CDCl_3$) δ, 1.46 (d, 3H, $J_{18,19} = 6.8$ Hz, C-18H), 1.8–2.22 (m, 2H, C-14H), 3.01–3.44 (m, 2H, C-3H), 3.78 (m, 1H, C-15H), 4.25 (d, 1H, $J_{6\alpha,6\beta} = 17.8$ Hz, (C-6Hα), 4.51 (d, 1H, $J_{6\beta,6\alpha} = 17.8$ Hz, C-6 Hβ), 5.25 (br. q, 1H, $J_{19,18} = 6.8$ Hz, C-19H), 7.03–7.43 (m, 4H, Ar-H), 7.85 (br. s, 1H, NH).

Fraction F_2 (20 g) was loaded on a silica column (750 g) which was eluted with increasing polarities of mixtures of petroleum ether, $CHCl_3$, EtOAc and MeOH. The fractions obtained on elution with petroleum ether: $CHCl_3$ (2:8) consisted of a mixture of two alkaloids. These fractions were combined, evaporated and subjected to preparative TLC (petroleum ether, acetone, aqueous ammonia, 7:3:0.05). The slower moving major alkaloid (25 mg) was identified as conoflorine (**4**).

***Conoflorine* (4)**

UV (MeOH): λ_{max}, 229, 285, 292 nm; IR ($CHCl_3$): ν, 1570, 1625 cm^{-1}; MS: *m/z* (%) = 296 (48), 267 (10), 249 (18), 211 (12), 157 (38), 143 (30), 140 (100), 122 (35); ^{1}H-NMR ($CDCl_3$): δ, 0.74 (t, 3H, $J_{18,19}$ = 7.5 Hz, C-18H), 1.14 (q, 2H, C-19H), 4.18 (m, 1H, C-14H), 7.03–7.46 (m, 4H, Ar-H), 7.78 (br. s, 1H, N$\underline{H}$).

Further elution of the same column with $CHCl_3$: EtOAc (9 : 1) afforded another alkaloid which was purified by preparative TLC on silica gel, (petroleum ether, EtOAc, 1 : 1). This alkaloid (12 mg) was identified as 19-epi-voacangarine (5).

***19-epi-Voacangarine* (5)**

UV (MeOH): λ_{max}, 224, 282, 298 nm; IR ($CHCl_3$): ν, 1730, 1625, 1578 cm^{-1}; MS: *m/z* (%) = 384 (49), 369 (20), 366 (57), 340 (9), 325 (5), 307 (12), 244 (27), 184 (38), 124 (33), 122 (11); ^{1}H-NMR ($CDCl_3$): δ, 1.28 (d, 3H, $J_{18,19}$ = 6.4 Hz, C-18H), 4.03 (m, 1H, C-19H), 3.73 (s, 3H, $COOC\underline{H}_3$), 3.85 (s, 3H, Ar-$OC\underline{H}_3$), 6.62 (dd, 1H, $J_{11,12}$ = 8.0 Hz, $J_{11,9}$ = 2.4 Hz, C-11H), 6.90 (d, 1H, $J_{9,11}$ = 2.4 Hz, C-9H), 7.15 (d, 1H, $J_{12,11}$ = 8 Hz, C-12H), 8.71 (br. s, 1H, NH).

References

(1) Perry, Z. M. (1895) Medicinal Plants of East and South East Asia, pp. 31–32, MIT Press, Cambridge.
(2) Gorman, M., Neuss, N., Cone, N. J., Deyrup, J. A. (1960) J. Am. Chem. Soc. 82, 1142.
(3) Walls, F., Collera, O., Sandoval, A. (1958) Tetrahedron 2, 173.
(4) Joule, J. A., Monteiro, H., Durham, L. J., Gilbert, B., Djerassi, C. (1965) J. Chem. Soc. 4773.
(5) Dugan, J. J., Hesse, M., Renner, V., Schmid, H. (1967) Helv. Chim. Acta 50, 60.
(6) Poisson, J., Puisieux, F., Miet, C., Patel, M. B. (1965) Bull. Soc. Chim. Fr. 3349.
(7) Wenkert, E., Cochran, D. W., Gottlieb, H. E., Hagaman, E. W., Filho, R. B., Matos, F. J. de A., Madruga, M. I. L. M. (1976) Helv. Chim Acta 59, 2437.

HPLC of Glycoalkaloids from *Solanum sodomaeum*

Bill E. Cham[1,2] and Linda Wilson[3]

Received: May 14, 1986

Abstract: Glycoalkaloids were extracted from plant material of *Solanum sodomaeum* L. High-performance liquid chromatographic studies indicated that several glycoalkaloids were present. The sugar moiety of the glycoalkaloids consisted of a mixture of glucose, rhamnose, and galactose. Mass spectral analyses showed that all the glycoalkaloids contained solasodine. Solasonine and solamargine were identified and were present at similar concentrations, representing 67 % of the total extracted glycoalkaloids. The extraction yield of the glycoalkaloids from plant material, expressed as g glycoalkaloid/100 g wet weight, was highest for ripe fruit (0.83 ± 0.11), followed by unripe fruit (0.45 ± 0.08), leaves (0.14 ± 0.01), and stems (< 0.04) with progressively decreasing yields. Over a twofold difference in glycoalkaloid yield was observed when ripe fruit of two different districts was extracted.

Introduction

Many well-known alkaloids currently used in Medicine are extracted from plants. B-solamarine, a glycoalkaloid extracted from *Solanum dulcamara* is a tumour inhibitor (1, 2). Most recently, it was also shown that glycoalkaloids extracted from the fruit of another *Solanum* species namely *Solanum sodomaeum* L. possessed antineoplastic activity both in mice (3) and in humans[4]. In this paper, we report the extraction, identification and purification of these glycoalkaloids from *Solanum sodomaeum* L.

Material and Methods

Plant materials

S. sodomaeum L. also known as *S. hermannii* Dunal is native to South Africa and the Mediterranean basin. It was introduced to Australia in 1801. The botanical description has already been documented (4). The fruit, leaves, and stems of *S. sodomaeum* L. used for this study were collected from wild plants in the Brisbane district.

Chemicals

Solasodine, solasonine, and solamargine were kindly supplied by Dr Jay Mann, Applied Biochemistry Division, DSIR, Lincoln, New Zealand. Fig. 1 illustrates the chemical structures of solamargine (1) (5) and solasonine (2) (6). All other chemicals were of high performance liquid chromatography (HPLC) grade for the HPLC studies and of analytical reagent grade for the extraction studies.

Extraction of glycoalkaloids from Solanum sodomaeum

The extraction procedure was a modification of that of Kuhn and Löw (7). Plant material for *S. sodomaeum* was coarsely ground with two volumes (w/v) of 3 % aqueous acetic acid in a Waring blender. The mixture was diluted with another two volumes of 3 % aqueous acetic acid and was then shaken for 18–20 h at room temperature, then filtered through muslin. (A minimum of 4 h was required for complete extraction of the glycoalkaloids). Two litre aliquots of the filtrate were heated to 50° C with continuous stirring and then concentrated ammonia solution was added until the pH reached 9–10 (approximately 50 ml/litre). The solution was maintained at 50° C for a further 5 min, allowed to cool, and then centrifuged at 2000 g for 15 min. The supernatant was discarded and the precipitate was dissolved in 1 litre 3 % aqueous acetic acid. The solution was centrifuged as above and the supernatant heated to 50° C with continuous stirring. The glycoalkaloids were reprecipitated on addition of concentrated ammonia solution until the pH reached 9–10. The solution was maintained at 50° C for a further 5 min as before, allowed to boil, cooled, then centrifuged as above. The pre-

---

[1] Department of Medicine, University of Queensland, Clinical Sciences Building, Royal Brisbane Hospital, Queensland, 4029, Australia.
[2] Address for correspondence.
[3] Marist Brothers College, Fernberg Road, Rosalie, Queensland, Australia
[4] [illegible] preparation.